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Association of PAH–DNA Adducts in Peripheral White Blood Cells with Dietary Exposure to Polyaromatic Hydrocarbons

by Nathaniel Rothman,[1] Miriam C. Poirier,[2] Robert A. Haas,[3] Adolfo Correa-Villasenor,[4] Patrick Ford,[5,6] John A. Hansen,[4] Tara O'Toole,[5,7] and Paul T. Strickland[5]

Previous investigations suggest that dietary sources of polycyclic aromatic hydrocarbons (PAHs) contribute to the PAH-DNA adduct load in peripheral white blood cells (WBCs). In the current study, we measured PAH–DNA adducts by enzyme-linked immunosorbent assay in WBCs obtained from 47 California wildland (forest) firefighters at two time points (early and late) during an active forest fire season. PAH–DNA adduct levels were not associated with recent firefighting activity, but were positively associated with frequency of charbroiled food consumption in the previous 2 weeks. In addition, adduct levels declined with time since last ingestion of charbroiled food. These studies indicate that recent consumption of charbroiled food contributes to the PAH–DNA adduct load in peripheral WBCs.

Introduction

Polycyclic aromatic hydrocarbons (PAHs) are carcinogenic compounds produced by incomplete combustion of organic material. Humans are exposed to these compounds from a wide variety of occupational, environmental and dietary sources. Measurement of PAH–DNA adducts in peripheral white blood cells (WBCs) has been proposed as a method for assessing exposure to these compounds. Previous studies suggest that this biomarker reflects both occupational (*1,2*) and dietary PAH exposure (*3,4*).

Wildland firefighters may have nearly continuous exposure to PAHs (*5*) for several weeks at a time while fighting large forest fires. The purpose of this study was to investigate the association between PAH–DNA adducts and occupational and dietary PAH exposures in a population of wildland firefighters.

[1]Epidemiology and Biostatistics Program, National Cancer Institute, Bethesda, MD 20892.
[2]Chemical and Physical Carcinogenesis Program, National Cancer Institute, Bethesda, MD 20892.
[3]California Department of Health Services, Berkeley, CA 94704.
[4]Department of Epidemiology, The Johns Hopkins University School of Hygiene and Public Health, Baltimore, MD 21205.
[5]Department of Environmental Health Sciences, The Johns Hopkins University School of Hygiene and Public Health, Baltimore, MD 21205.
[6]Present address: Occupational Health/Preventive Medicine Section, Naval Hospital, Philadelphia, PA.
[7]Present address: U.S. Congressional Office of Technology Assessment, Washington, DC.
[8]Address reprint requests to P. T. Strickland, Department of Environment Health Services, The Johns Hopkins University School of Hygiene and Public Health, 615 N. Wolfe Street, Room 2708, Baltimore, MD 21205.

Methods

Forty-seven nonsmoking firefighters (37 male, 10 female), 18–45 years old, were evaluated in July and September of 1988, the early and late part of the northern California wildland fire season. Informed consent was obtained from all subjects, who were stationed at several of the most active wildland fire stations in the region. At the beginning and end of the 8-week study period, a questionnaire requesting information on demographics, work practices, frequency of charbroiled (CB) food consumption in the previous 2 weeks, and time since CB food was last consumed was self-administered, and 40 mL of blood were obtained.

DNA extracted from WBCs was analyzed for PAH–DNA adduct content by enzyme-linked immunosorbent assay (ELISA) (*6*) using a standard benzo[*a*]pyrene diol epoxide I (BPDE)–DNA modified in the same range as the biological samples (4.4 fmole/μg DNA) (*7*). The ELISA employed rabbit anti-BPDE–DNA (antibody 33, diluted 1:70,000), which recognizes several PAH–DNA adducts. These PAHs are present in roughly similar porportions in forest fire smoke (unpublished data) and charbroiled food (*8*). The lower limit of detection was 0.04 fmole adduct/μg DNA at 15% inhibition. Samples with nondetectable levels (39 of 94) were assigned a value of 0.02 fmole adduct/μg DNA.

Due to the skewed nature of adduct level distribution, the relationship between PAH–DNA adduct levels, firefighting activity, and CB food intake was examined by linear regression using adduct level rank (*9*) as the outcome variable and logistic regression using adduct levels categorized into $\leq$ 0.2 fmole/μg DNA

Table 1. PAH-DNA adduct levels and consumption of charbroiled food.

Attribute	n[a]	PAH-DNA adduct level, fmole/μg DNA Mean (SD)	Median (range)
Times ate charbroiled food in past 2 weeks[a]			
0	24	0.06 (0.07)	0.02 (0.02-0.24)
1-2	37	0.10 (0.12)	0.06 (0.02-0.6)
3-5	28	0.13 (0.13)	0.08 (0.02-0.42)
>5	5	0.21 (0.17)	0.21 (0.02-0.38)
Weeks since last ate charbroiled food[c]			
≤ 1 week	45	0.13 (0.14)	0.08 (0.02-0.6)
> 1 week	39	0.07 (0.07)	0.02 (0.02-0.24)
Unknown	10	—	—

[a]Total samples = 94.
[b]Association between adduct level and frequency of charbroiled food consumption tested by linear regression on adduct level rank (p = 0.016)
[c]Association between adduct level and weeks since last ate charbroiled tested by linear regression on adduct level rank (p = 0.03).

or > 0.2 fmole/μg DNA. [Previous studies have demonstrated that individuals without occupational (*2,4,10*) or marked dietary PAH exposure (*2*) generally have peripheral WBC PAH-DNA adduct levels below 0.2 fmole/μg DNA.] Potential confounders evaluated included age, race, sex, passive exposure to cigarette smoke, diesel exhaust exposure, years as a firefighter, use of a bandana for respiratory protection, and alcohol and coffee consumption.

Early and late season data were analyzed as a single study using the method of Liang and Zeger (*11*), which takes into account the possible correlation between repeat outcome measurements made on each individual. Two-tailed p-values were calculated throughout.

Results

Frequency of CB food consumption in the previous 2 weeks ranged from zero to more than eight times (median: one to two times). PAH-DNA adduct levels were positively associated with CB food consumption (Table 1). Median duration since individuals last ingested CB food was 6 days (range: 1 to > 30 days). Adduct levels were significantly higher in individuals who had ingested CB food one or more times within the previous week (Table 1).

Individuals who consumed CB food more than twice in the previous 2 weeks had a 4-fold increased risk of having elevated adduct levels (> 0.2 fmole/μg DNA) compared to individuals consuming CB food two or fewer times (Table 2). Individuals who ingested CB food within the previous week had a 4-fold increased risk of having elevated adduct levels compared to individuals who last ingested CB food more than 1 week earlier (Table 3).

None of the above associations were significantly altered after adjustment for potential confounding variables and firefighting activity. There was no association between firefighting activity and PAH-DNA adduct level (Rothman et al., manuscript in preparation).

Table 2. Association of elevated PAH-DNA adduct levels with frequency of charbroiled food consumption.[a]

Times ate charbroiled food in last 2 weeks	PAH-DNA adduct level > 0.2 fmole/μg DNA	≤ 0.2 fmole/μg DNA	Total
> 2 times	10	23	33
≤ 2 times	6	55	61
Total	16	78	94

[a]Odds ratio = 4.1; 95% C.I. = 1.4,12.03; p = 0.009. Crude odds ratio, 95% C.I., and p-value determined by logistic regression.

Table 3. Association of elevated PAH-DNA adduct levels with weeks since last consumed charbroiled food.[a]

Weeks since last ate charbroiled food	PAH-DNA adduct level > 0.2 fmole/μg DNA	≤ 0.2 fmole/μg DNA	Total
≤ 1 week	11	34	45
> 1 week	3	36	39
Total	14	70	84[b]

[a]Odds ratio = 3.9; 95% C.I. = 1.1, 13.9; p = 0.034. Crude odds ratio, 95% C.I., and p-value determined by logistic regression.
[b]Total samples do not add to 94 because data were missing for several study subjects.

Dicussion

Blood samples from 47 California wildland firefighters obtained early and late in the 1988 forest fire season were analyzed for WBC PAH-DNA adduct levels by ELISA. Adduct levels were positively associated with frequency of CB food consumption and inversely associated with time since CB food was last consumed. This association is in accordance with our previously published findings from a cross-sectional study of urban firefighters and matched controls (*3*) and a controlled feeding study in human volunteers (*4*). Furthermore, the observation that only recent CB food consumption is associated with adduct levels is consistent with the relatively short half-life (< 24 hr) of a substantial portion of peripheral WBCs (*12*).

Although PAHs are ubiquitous in the diet, CB food has been consistently shown to contain some of the highest PAH levels in commonly consumed food items (*13*). Our data also suggest that CB food represents one of the major sources of dietary PAH intake for populations that frequently prepare food by charbroiling. Further exploration of the association between diet and PAH biomarkers should, however, attempt to evaluate exposure to all major PAH dietary sources.

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